International Journal of Nanomedicine

Dovepress
Taylor & Francis Group

Open Access Full Text Article

ORIGINAL RESEARCH

Formulation and Characterization of *Sonchus arvensis* L. Nanosuspension for Enhanced Antioxidant and Lipid-Lowering Activities

Mohamed Mahmud E Aldeeb[1–3], Gofarana Wilar[4], Cecep Suhandi[1,2], Ahmed Fouad Abdelwahab Mohammed[5], Ali El-Rayyes[6], Khaled M Elamin[7], Nasrul Wathoni[2]

[1]Doctoral Program of Pharmacy, Faculty of Pharmacy, Universitas Padjadjaran, Jatinangor, 45363, Indonesia; [2]Department of Pharmaceutics and Pharmaceutical Technology, Faculty of Pharmacy, Universitas Padjadjaran, Jatinangor, 45363, Indonesia; [3]Department of Pharmaceutics, Faculty of Pharmacy, Elmergib University, Alkhoms, 40414, Libya; [4]Department of Pharmacology and Clinical Pharmacy, Faculty of Pharmacy, Universitas Padjadjaran, Jatinangor, 45363, Indonesia; [5]Department of Pharmaceutics, Faculty of Pharmacy, Minia University, Minia, 61519, Egypt; [6]Center for Scientific Research and Entrepreneurship, Northern Border University, Arar, 73213, Saudi Arabia; [7]Graduate School of Pharmaceutical Sciences, Kumamoto University, Kumamoto, 862-0973, Japan

Correspondence: Nasrul Wathoni, Department of Pharmaceutics and Pharmaceutical Technology, Faculty of Pharmacy, Universitas Padjadjaran, Jatinangor, 45363, Indonesia, Tel +62-22-842-888-888, Email nasrul@unpad.ac.id

Purpose: This study aimed to develop a nanosuspension formulation of *Sonchus arvensis* L. folium to enhance its antihyperlipidemic effectiveness by optimizing its particle size, stability, and pharmaceutical availability.

Methods: Nanosuspensions were prepared using an antisolvent technique, with Tween 80 as the stabilizing agent. The formulations were characterized by particle size analysis, zeta potential, and polydispersity index (PDI) to evaluate their stability and uniformity. Transmission electron microscopy (TEM) confirmed nanoparticle formation, and stability assessments were conducted over 28 days under refrigerated conditions. In vitro release studies were conducted to assess sustained drug release, and antioxidant activity and lipid-lowering efficacy were compared to those of simvastatin.

Results: The NS-Sa-4:1 formulation exhibited optimal properties, including an average particle size of 12.9 nm, zeta potential of −12.5 mV, and PDI of 0.317, indicating a stable and uniform nanosuspension. TEM images revealed well-dispersed nanoparticles, and stability tests demonstrated the retention of these properties for up to 28 days. In vitro release studies showed sustained drug release, with 92.51% released within 24 h, surpassing the crude extract and other formulations. NS-Sa-4:1 also demonstrated enhanced antioxidant activity and significant lipid-lowering effects compared with simvastatin.

Conclusion: The nanosuspension of *Sonchus arvensis* L. folium, particularly NS-Sa-4:1, shows significant potential as an effective antihyperlipidemic therapy with improved stability, pharmaceutical availability, and therapeutic efficacy. Future research should focus on pharmacokinetics and in vivo studies to further validate these results.

Keywords: *Sonchus arvensis* L., nanosuspension, antioxidant, antihyperlipidemic

Introduction

Hyperlipidemia is a medical disorder characterized by elevated lipid or fat levels in the bloodstream. These lipids, primarily cholesterol and triglycerides, play vital roles in maintaining numerous physiological functions. However, when present in excessive amounts, they can contribute to a heightened risk of cardiovascular conditions, including atherosclerosis, heart attacks, and stroke. Various medications have shown promise in lowering serum cholesterol. Ezetimibe combined with statins has yielded moderate benefits, whereas bempedoic acid has demonstrated a 15% LDL cholesterol reduction in trials. For patients with persistently high lipid levels despite statin therapy, FDA-approved PCSK9 inhibitors are now recommended. Newer options, such as inclisiran, an siRNA therapy administered biannually, show the potential to improve compliance, especially when used with pelacarsen, which targets lipoprotein (a). Additionally, Volanesorsen and its successor olezarsen

Received: 21 December 2024
Accepted: 31 March 2025
Published: 27 April 2025

target chylomicrons to reduce triglyceride levels, and ANGPTL3 inhibitors, such as evinacumab, have been approved for familial hypercholesterolemia, highlighting significant advances in lipid-lowering treatments.[1]

Herbal remedies are viable alternatives to commercially available cholesterol-lowering medications, particularly when cardiovascular illnesses are the leading cause of mortality.[2,3] CVD risk stratification should be considered when selecting the appropriate medications. The risk is determined by age, severity, and presence of additional personal or familial CVD risk factors. Lifestyle changes are recommended, with a particular incidence of dietary treatment for 3–6 months before pharmacological therapy reassessment and decision-making.[4,5]

In recent years, there has been a growing interest in the therapeutic potential of natural products and plant-based compounds.[6,7] Natural compounds derived from medicinal plants have demonstrated significant promise in treating various ailments due to their bioactive constituents, which offer a wide range of pharmacological activities with minimal side effects compared to synthetic drugs. These natural products are often rich in polyphenols, flavonoids, and alkaloids, which contribute to their therapeutic efficacy. *Sonchus arvensis* L., commonly known as field sow thistle, is a vascular plant belonging to the phylum Tracheophyta and the class Magnoliopsida (dicotyledons). It is systematically classified under the order Asterales, within the family Asteraceae (daisy family), and the genus Sonchus. It is a medicinal plant that has been used in traditional medicine because of its diverse pharmacological properties, including antioxidant, anti-inflammatory, and hypolipidemic effects.[8,9] The plant has been reported to exhibit additional pharmacological benefits, such as antimicrobial, hepatoprotective, nephroprotective, and antidiabetic properties. The presence of bioactive compounds, including flavonoids, sesquiterpene lactones, and phenolic acids, enhances its medicinal value, making it a potential candidate for novel drug formulations.

These characteristics make *Sonchus arvensis* L. a promising candidate for developing innovative drug delivery systems. Unlike many other herbal formulations, which suffer from issues such as poor solubility, rapid degradation, and inconsistent bioavailability, *Sonchus arvensis* L. has been recognized for its high stability and potent bioactivity, making it particularly well-suited for advanced pharmaceutical applications. Building on the therapeutic potential of *Sonchus arvensis* L.[10]

The development of poorly soluble drugs presents a major challenge in the pharmaceutical industry because of their limited solubility and bioavailability, which can significantly reduce their therapeutic efficacy.[11,12] One promising strategy for addressing these limitations is the formulation of nanosuspensions.[13] Nanosuspensions are colloidal dispersions of submicron-sized drug particles stabilized by surfactants, which enhance the solubility, stability, and bioavailability of poorly soluble drugs.[14] By reducing the particle size to the nanometer range (<100 nm),[15] nanosuspensions increase the surface area available for dissolution, which improves both the rate and extent of drug absorption. Additionally, these formulations can be designed to achieve controlled and sustained drug release, thereby improving patient compliance and therapeutic outcomes.[16–19] *Sonchus arvensis* L.-based nanosuspensions offer several advantages over other herbal nanosuspensions. First, its strong antioxidant and anti-inflammatory properties provide synergistic therapeutic effects, enhancing the overall efficacy of the nanosuspension. Second, its diverse range of bioactive compounds allows for multifaceted therapeutic applications, targeting multiple pathways in disease treatment. Furthermore, the inherent stability and low toxicity profile of *Sonchus arvensis* L. make it a superior choice for nanosuspension formulations, ensuring better safety and efficacy for long-term use. These advantages underscore its potential as a groundbreaking component of modern herbal nanomedicine. This study aimed to formulate and evaluate a nanosuspension-based drug delivery system utilizing *Sonchus arvensis* L.

Materials and Methods

Materials

Sonchus arvensis L. folium were harvested by the farmer from Cikanyere village, Sukanyere subdistrict, Cianjur, Indonesia. A voucher specimen (Herbarium No: 642) was deposited at the Herbarium Jatinangoriense, Biosystematics and Molecular Laboratory, Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Padjadjaran. The materials used in this investigation were distilled water, 2,2-diphenyl-1-picrylhydrazyl (DPPH), Lieberman-Burchard reagent, phosphate buffer saline (PBS), acetic anhydride, aluminum chloride ($AlCl_3$), anhydrous

sodium sulfate, chloroform, 70% ethanol, ferric chloride ($FeCl_3$), gallic acid, hydrochloric acid (HCl), potassium dihydrogen phosphate (KH_2PO_4), potassium acetate (CH_3CO_2K), polysorbate 80 (Tween 80®), quercetin, sodium carbonate (Na_2CO_3), sodium hydroxide (NaOH), Folin-Ciocalteu reagent, and Dragendorff's reagent, all sourced from Sigma-Aldrich Co., St. Louis, MO, USA.

Methods

This study was conducted in four stages. Initially, an extract of *Sonchus arvensis* L. was obtained and subjected to phytochemical screening. This was followed by the formulation of a nanosuspension and its characterization, including particle size analysis (PSA), polydispersity index (PI), and zeta potential measurements. Short-term stability evaluation was conducted for 28 days, after which transmission electron microscopy (TEM) was used for further analyses. The study concluded with an in vitro release assessment and evaluation of the antioxidant and antihyperlipidemic properties of the prepared nanosuspensions.

Extraction and Phytochemical Screening of *Sonchus arvensis* L.

The extract of *Sonchus arvensis* L. was prepared using the maceration technique. Initially, 1.5 kg of sifted *Sonchus arvensis* L. powder was added. Leaves prepared using a no. 40 mesh sifter were accurately weighed. This powder was then extracted with 16 liters of 70% ethanol solvent over three days. The extraction process was organized into three separate maceration vessels with intermittent stirring to prevent saturation. The resulting filtrate was concentrated using a rotary evaporator at 60°C. Subsequently, the concentrated extract was evaporated in a water bath to obtain a viscous extract was achieved.[20]

$$\text{Yield}\ (\%\,\text{w/w}) = (\text{viscous extract weight}/\text{dry } \textit{Sonchus arvensis}\ \text{L. weight}) \times 100\%.$$

Phytochemical Screening of *Sonchus arvensis* L. Extract

In this study, phytochemical screening was performed to determine the presence of alkaloids, flavonoids, tannins, and saponins in the extracts. Each extract exhibited positive indicators, including sediment and foam formation and color changes.

Measurement of Total Flavonoid Content (TFC)

The Total Flavonoid Content (TFC) was determined following the guidelines outlined in the Indonesian Herbal Pharmacopeia 2nd edition 2017. The procedure involved preparing both the sample extract and standard quercetin solution. For the sample preparation, approximately 0.2 g of the extract was accurately weighed and placed into an Erlenmeyer flask, followed by the addition of 25 mL of ethanol P. The mixture was stirred for 30 min using a magnetic stirrer to ensure thorough extraction. Subsequently, the solution was filtered into a 25-mL volumetric flask, and ethanol P was added to reach the final volume.

For the preparation of the standard solution, approximately 10 mg of quercetin was weighed and dissolved in ethanol P in a 25-mL volumetric flask. A series of dilutions were prepared to obtain standard solutions with concentrations of 100, 75, 50, and 25 mg/mL, which were used to generate the calibration curve.

In the test procedure, 0.5 mL of both the test and standard solutions were separately pipetted into vials. To each vial, 1.5 mL of ethanol P, 0.1 mL of 10% aluminum chloride P, 0.1 mL of 1 M sodium acetate, and 2.8 mL of water were added. The mixture was then shaken and allowed to stand for 30 min at room temperature to allow for color development. The absorbance of each solution was measured at the maximum absorption wavelength, and a blank solution prepared without the addition of aluminum chloride was used as a reference. Finally, a calibration curve was constructed using standard quercetin solutions, and the flavonoid concentration in the test sample was determined based on this curve.

Measurement of Total Phenolic Content (TPC)

The Total Phenolic Content (TPC) was determined using a standardized procedure to ensure accuracy and reproducibility. The process began with the preparation of the sample extract, where approximately 0.2 g of the extract was accurately weighed and placed in an Erlenmeyer flask. To facilitate extraction, 25 mL of methanol P was added, and the mixture was stirred for 30 min using a magnetic stirrer. Following extraction, the solution was filtered into a 25-mL volumetric flask, and methanol P was added to obtain the final volume.

For the preparation of the standard solution, approximately 10 mg of gallic acid was weighed and dissolved in methanol P in a 25-mL volumetric flask. A series of dilutions were prepared to obtain standard solutions with concentrations of 100, 80, 40, 20, and 10 µg/mL, which were subsequently used to generate the calibration curve.

In the test procedure, 1 mL of both the test and standard solutions were separately pipetted into vials. To each vial, 5.0 mL of diluted Folin-Ciocalteu reagent (7.5% in water) was added, and the mixture was allowed to stand for 8 min to ensure complete reaction. Subsequently, 4.0 mL of 1% NaOH was added, and the solutions were incubated for 1 h to allow color development. The absorbance of each solution was measured at the maximum absorption wavelength of approximately 730 nm. A blank solution prepared using the same procedure but without adding the test sample was used as a reference. Finally, a calibration curve was constructed using standard gallic acid solutions, and the phenolic content in the test sample was determined based on this curve.[21]

Formulation of Nanosuspension

The nanosuspension was prepared using the nanoprecipitation method, as shown in Figure 1, by combining the organic and aqueous phases, as presented in Table 1. *Sonchus arvensis* L. extract (0.25 g) was dissolved in 10 mL of ethanol

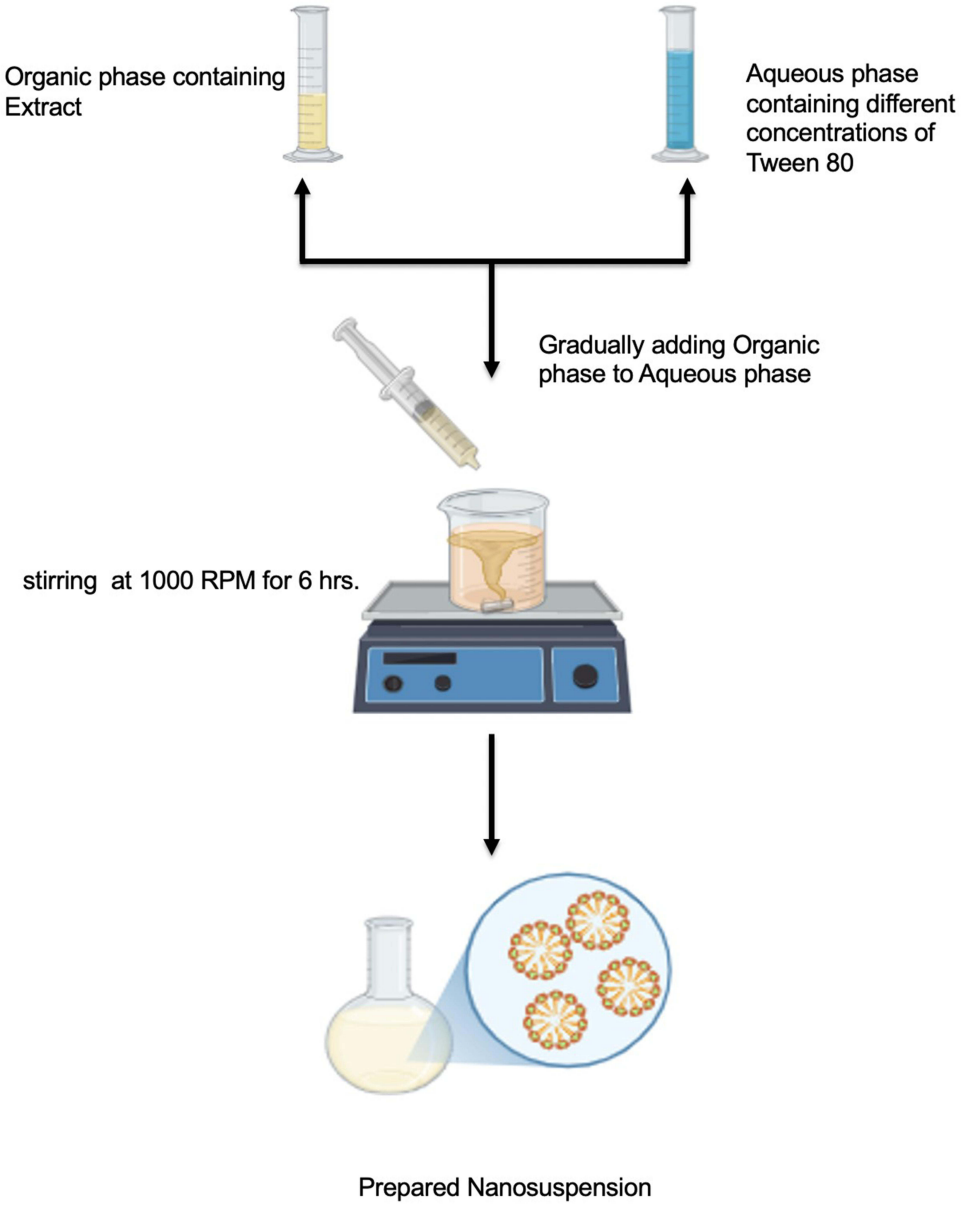

Figure 1 Nanoprecipitation method for *Sonchus arvensis* nanosuspension preparation.

Table 1 Formulation of Nanosuspension Tween 80 to Extract Ratios

No. of Formula	Amount of Stabilizer g	Amount of Extract g	Amount of Organic Solvent mL	Amount of Anti-Solvent mL
NS-Sa-1:1	0.25 g	0.25 g	10 mL	100 mL
NS-Sa-2:1	0.5 g	0.25 g	10 mL	100 mL
NS-Sa-4:1	1 g	0.25 g	10 mL	100 mL

(organic phase), and 0.25, 0.5, or 1 g of the stabilizer (Tween 80) was dissolved in 100 mL of distilled water (aqueous phase). The resulting organic layer was gradually (1mL/min) poured into the aqueous phase using a syringe with constant stirring at 1000 rpm for 6 h. at room temperature.[22–24]

Characterization of Utilized Nanosuspension

Particle size analysis (PSA) of the *Sonchus arvensis* L. formulations was conducted by assessing the z-average diameter and measuring the polydispersity index (PI) using photon correlation spectroscopy (PCS) with a Zetasizer (NanoSizer 3000, Malvern Instruments, Malvern, UK) at a scattering angle of 90°.[25] The measurements were performed in a cell with a width of 1 cm at a controlled temperature of 25.0 ± 0.1 °C. Additionally, the zeta potential (ZP) of the nanosuspension was measured in a capillary cell using the same instrument, which employed the Helmholtz–Smoluchowski equation to convert the observed particle electrophoretic mobility into zeta potential values. The zeta potential reflects the surface charge of the nanoparticles and serves as an indicator of their physical stability.[26]

Transmission Electron Microscopy (TEM) Analysis

The surface morphologies of the prepared nanosuspensions and extracts were examined using transmission electron microscopy (TEM).[27] A small quantity of nanosuspensions and extracts was carefully deposited dropwise onto a specially designed copper grid for TEM analysis. Subsequently, the copper grid was placed in an oven at 45 °C to ensure complete drying of the sample. The dried samples were analyzed using an HT7700 TEM (Hitachi, Tokyo, Japan). Imaging was conducted at a voltage of 100 kV with a magnification of ×100,000, and video recordings were captured for 0.1 s. The resulting images were processed using ImageJ 2.0 software.[28,29]

In vitro Release Studies of *Sonchus arvensis* L. Extract and Formulated Nanosuspension

An in vitro release study of the extracts and three optimized nanosuspensions was conducted using the dialysis bag technique (Figure 2).[30] The release experiments were performed at an intestinal pH of 7.4, utilizing phosphate-buffered saline (PBS) at the same pH level. In this procedure, pre-weighed samples of the extract and nanosuspensions were placed into pre-soaked dialysis bags with a molecular weight cut-off (MWCO) of 12–14 kDa (Carolina Dialysis Tubing, Burlington, NC). The release process was carried out in 100 mL of PBS at pH 7.4, maintained at a temperature of 37 ± 0.5°C with continuous stirring at 100 rpm using a magnetic stirrer. Samples were collected at predetermined time points (0, 1, 2, 3, 4, 5, 6, 8, and 24 h). Following each sampling, an equal volume of fresh medium was added to maintain sink conditions. The quercetin content in the collected samples was analyzed using a UV-Vis spectrophotometer at a wavelength of 345 nm.[24,30]

In vitro Antioxidant Activity Assay of Formulated Nanosuspensions and Crude Extract

The antioxidant properties of the nanosuspension formulations were evaluated using the DPPH scavenging assay, which measures the ability of antioxidant compounds to donate hydrogen atoms to neutralize the DPPH radicals. For the assay, various concentrations (50, 100, 150, 200, and 250 ppm) of each sample, including the *Sonchus arvensis* L. extract and its formulated nanosuspension, were prepared by dilution in 96% ethanol. A 1 mL aliquot of 0.2 mm DPPH solution was then added to 1 mL of each sample, followed by thorough mixing. After a 30 min incubation period in the dark, the absorbance was recorded at 517 nm using a UV-Vis spectrophotometer.[31] All experiments were performed in triplicates.

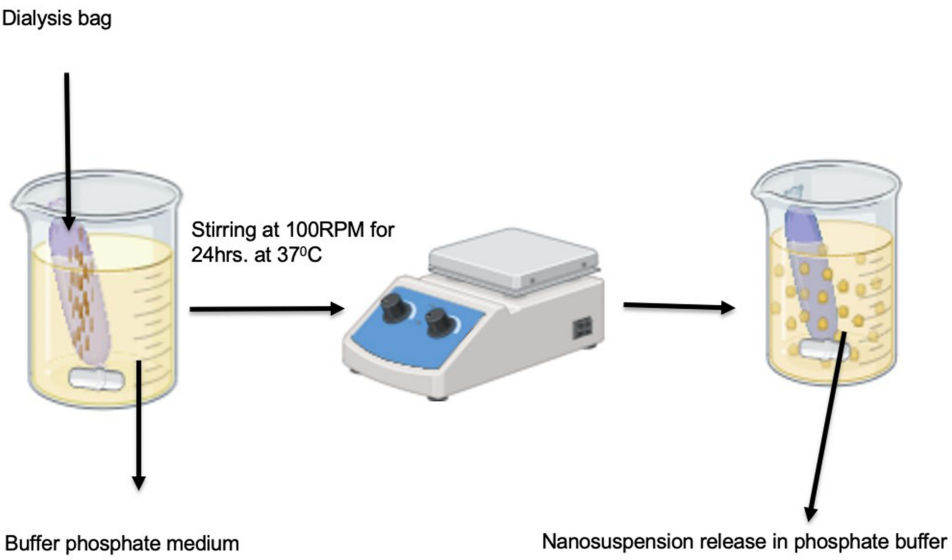

Figure 2 The in vitro release study using the dialysis bag technique.

Serial dilutions of ascorbic acid were used as positive controls. The percentage of DPPH radical-scavenging activity was calculated using the following equation:

$$\text{Inhibition}\,(\%) = (\text{absorbance of control} - \text{absorbance of sample})/\text{absorbance of control}) \times 100\%$$

In vitro Anti-Hyperlipidemic Studies of *Sonchus arvensis* L. Extract and Prepared Nanosuspensions

The anti-cholesterol activity of the extract and prepared nanosuspension was evaluated using a photometric method, wherein cholesterol was allowed to react with the Liebermann-Burchard reagent. The crude extract and nanosuspension formulations were tested across a concentration range of (15.625, 31.25, 62.5, 125, and 250 mg/L). A volume of 4 mL was collected and subsequently transferred to a capped test tube, to which 1 mL of a 1000 ppm cholesterol stock solution was added, shaken for 2 min, and 2 mL of the Liebermann-Burchard reagent was then added. The mixture was incubated for 15 min in the dark, the experiment was conducted in triplicate, and the colorimetric results were quantified using a UV-Vis spectrophotometer at a peak wavelength of 620 nm. A blank control was established using a solution of 5 mL chloroform combined with 2 mL of Liebermann-Burchard reagent. The negative control consisted of 1 mL of 1000 ppm cholesterol solution in 5 mL chloroform supplemented with 2 mL Liebermann–Burchard reagent. A positive control was prepared using simvastatin at concentrations of (15.625, 31.25, 62.5, 125, and 250 mg/L). The residual cholesterol concentration was determined from the absorbance data measured at 620 nm.[32,33] The percentage inhibition was calculated using the following formula:

$$\text{Inhibition}\,(\%) = (\text{absorbance of control} - \text{absorbance of sample})/\text{absorbance of control}) \times 100\%$$

Results

Sonchus arvensis L. Extraction

The yields obtained from the maceration method for the extraction of *Sonchus arvensis* L. are presented in Table 2. The resulting yield was 12.38% (w/w), meeting the specified standard of the Indonesian herbal pharmacopeia for *Sonchus arvensis* L. extract, which requires a minimum yield of 7.5%.[21]

Table 2 *Sonchus arvensis* Standardization Results

Parameters	Results	Requirements
Yield (%)	12.38%	Not less than 7.5%
Total Flavonoids Content	33.9 ±1.5 mg QE/g	Meets the requirements
Total phenolics Content	30.3 ±0.06 mg GAE/g	Meets the requirements

Phytochemical Screening of *Sonchus arvensis* L. Extract

The phytochemical composition of the *Sonchus arvensis* L. extract was assessed using different reagents to identify the presence of various secondary metabolites (Table 3). The observed visual changes in the extract following each treatment are shown in Figure 3.[21]

Measurement of Total Flavonoid Levels of *Sonchus arvensis* L. Extract

The total flavonoid content of the *Sonchus arvensis* L. extract is presented in Table 1. The extract was found to have a flavonoid concentration equivalent to 33.9 ± 1.5 mg of quercetin equivalents (QE) per gram of extract.[21]

Measurement of Total Phenol Levels of *Sonchus arvensis* L. Extract

The phenolic content of the *Sonchus arvensis* L. extract, as presented in Table 2, was determined to be 30.3 ± 0.06 mg GAE/g gallic acid.[21]

Characterization of Prepared Nanosuspensions

The prepared nanosuspensions were characterized in terms of particle size distribution, polydispersity index, zeta potential, and short-term stability (28 days).

Table 3 Phytochemical Screening of *Sonchus arvensis* Extract Results

Secondary Metabolites	Reagents	Result*
Alkaloids	Mayer Dragendorff	(+)
Phenolics	$FeCl_3$ 1%	(+++)
Tannins	$FeCl_3$ 1%	(+++)
Flavonoids	Concentrated HCl + Mg powder Amyl alcohol	(+++) (+++)
Tannins	Gelatin 1%	(+++)
Quinones	KOH 5%	(+++)
Saponins	Shaking	(+++)
Coumarins	NH4OH 10%+UV 366	(−)
Triterpenoids **Steroids**	Concentrated H2SO4 + acetic anhydride	(+) (−)
Monoterpenoids/sesquiterpenoids	Vanillin Sulphate 10%	(+)

Notes: *(+) indicates the weak presence of the compound, (+++) indicates the strong presence of the compound, and (−) indicates the absence of the compound.

https://doi.org/10.2147/IJN.S511602

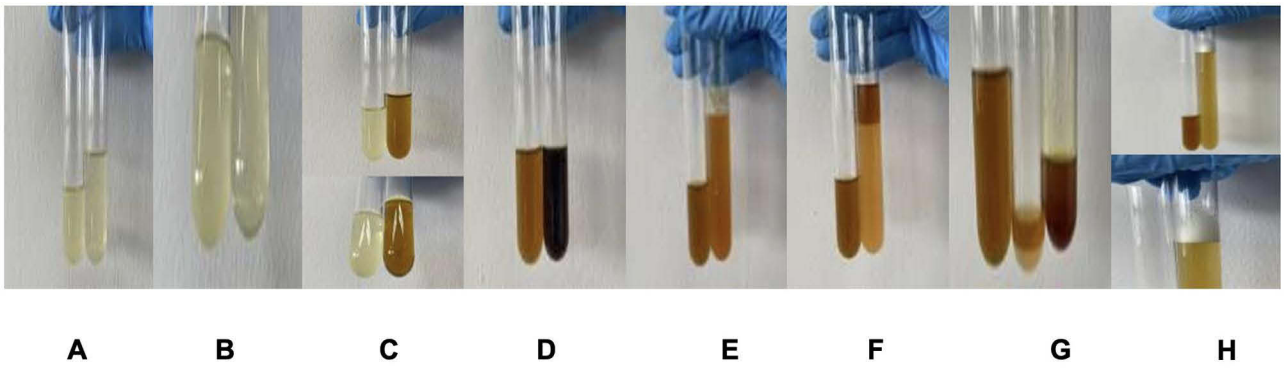

Figure 3 Phytochemical screening results. (**A** and **B**) Alkaloids forming white precipitate using Mayer reagent, (**C**) Dragendorff reagent forming an Orange-brown precipitate, (**D**) using FeCl3 1% forming a blue-black color for polyphenols, (**E**) Mg powder + 5 mL HCl 2N forming an Orange/red color for flavonoids; (**F**) Amyl alcohol reagent forming a yellow-red color for flavonoids (**G**) Gelatin 1% for tannins forming white precipitate or turbidity, (**H**) saponins after shaking yield persistent foam.

Particle Size Analysis

Initially, the particle size decreased with increasing Tween concentration, ranging from 13.133 nm for NS-Sa-1:1 to 11.533 nm for NS-Sa-4:1 (Table 4). This reduction reflects surfactant-induced stabilization, as higher Tween concentrations reduced the particle size. However, during the stability study, a slight increase in particle size was observed across all formulations. Specifically, after storage, the particle size of the NS-Sa-1:1 ratio increased to 14.23 nm, the NS-Sa-2:1 ratio increased to 13.13 nm, and the NS-Sa-4:1 ratio increased slightly to 12.9 nm. NS-Sa-4:1 exhibited the most stable particle size over time (Figure 4), indicating that a higher concentration of Tween could effectively limit particle growth.[34]

Polydispersity Index

In terms of PI, all formulations initially exhibited low values (0.145–0.161), suggesting a narrow and favorable particle size distribution. However, after 28 days of storage, the PI values increased across all formulations, likely reflecting a broadening of the size distribution due to particle aggregation. Specifically, the PI for NS-Sa-1:1 increased from 0.161 to 0.324 for NS-Sa-2:1, from 0.145 to 0.292 for NS-Sa-4:1, and from 0.146 to 0.317. Although NS-Sa-4:1 showed slightly improved stability, all ratios showed an increase in polydispersity, which may indicate minor instability during prolonged storage.

Zeta Potential

Zeta potential measurements initially varied with Tween concentration, with values decreasing from −6.26 mV at the NS-Sa-1:1 ratio to −15.033 mV at the NS-Sa-2:1 ratio, before rising slightly to −12.5 mV at the NS-Sa-4:1 ratio. More negative zeta potential values suggest greater electrostatic stability, and these values changed accordingly over the storage period. For NS-Sa-1:1, the zeta potential increased from −6.26 mV to −13.1 mV, implying improved stability. However, NS-Sa-2:1 decreased from −15.033 mV to −9 mV, indicating diminished electrostatic repulsion and a greater risk of aggregation (Figure 4). The NS-Sa-4:1 zeta potential remained stable at approximately −12.5 mV,[35] underscoring the effectiveness of higher Tween concentrations in maintaining electrostatic stability over time.

Table 4 Characterization of Prepared Nanosuspension (PSA, PI, Zeta Potential, n=3)

Tween 80 to Extract Ratios	PSA (nm)	Polydispersity Index (PI)	Zeta Potential (mV)
NS-Sa-1:1	14.23 ± 4.308	0.324 ± 0.102	−13.1± 5.115
NS-Sa-2:1	13.13 ± 1.106	0.292± 0.111	−9 ± 6.122
NS-Sa-4:1	12.9 ± 2.357	0.317 ± 0.079	−12.5 ± 1.915

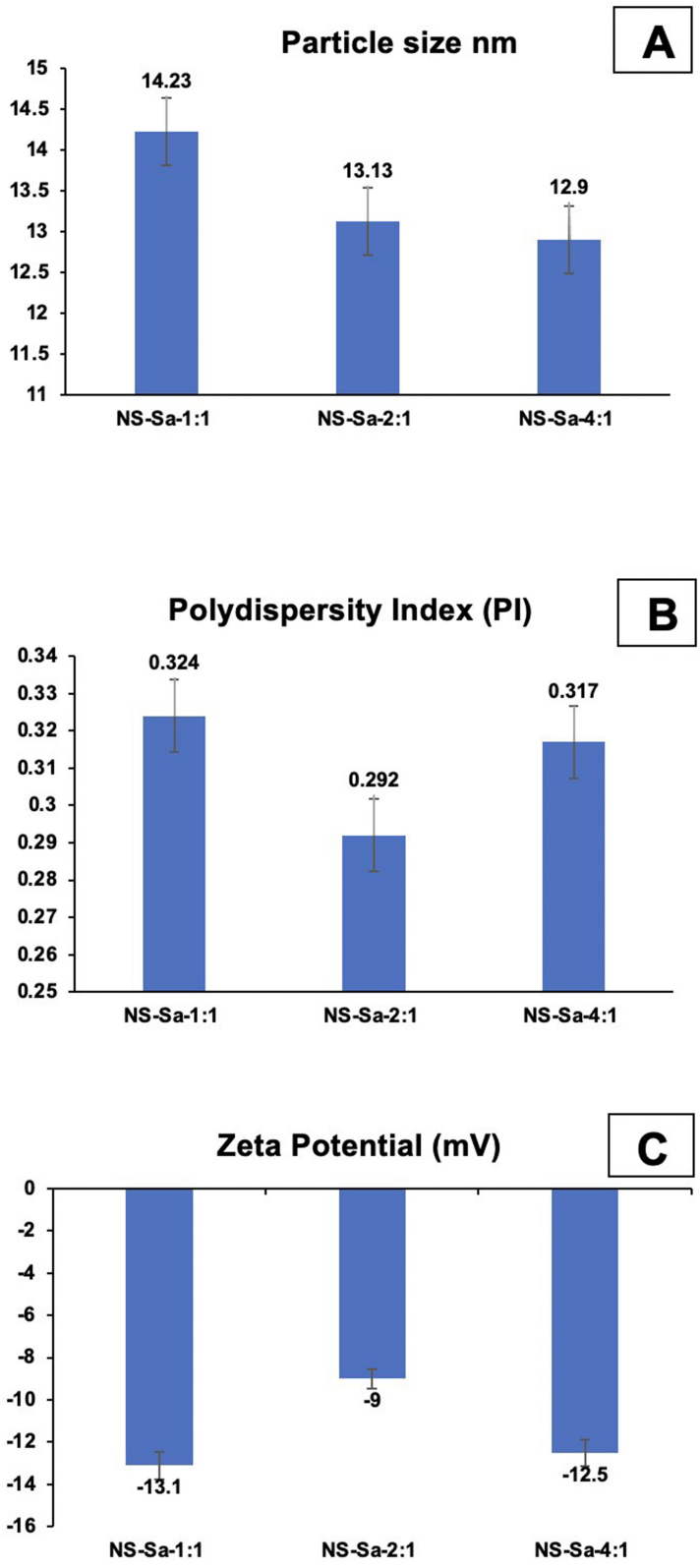

Figure 4 Stability studies of three formulations of nanosuspension (**A**) Particle size, (**B**) Polydispersity Index (PI), (**C**) zeta potential.

https://doi.org/10.2147/IJN.S511602

In the characterization and stability study (28 days), NS-Sa-4:1 demonstrated the most consistent stability across particle size, PI, and zeta potential. Overall, the particle sizes across all formulations remained within the nanoscale range (1–100 nm),[36] reaffirming their suitability for nanosuspension applications in drug delivery systems.

Transmission Electron Microscopy (TEM) Analysis

TEM analysis revealed that the raw *Sonchus arvensis* L. extract (Figure 5A) displayed an irregular and variable morphology, which is common in unprocessed plant extracts because of their natural diversity in particle size and shape. In contrast, (Figure 5B–D), representing the three different formulations, showed consistent spherical morphology, indicating that each formulation successfully produced a nanosuspension. This spherical shape suggests that the extract components were effectively transformed into nanoparticles, likely enhancing the stability and homogeneity of the formulations compared to the original extract.

Each formulation was prepared using varying concentrations of Tween 80, a surfactant that plays a crucial role in stabilizing nanoparticles by preventing particle aggregation. The consistent spherical morphology across all three formulations, regardless of the Tween 80 ratio, indicated that each formulation achieved a stable nanosuspension. This nanosuspension formation is promising for improving the solubility, stability, and pharmaceutical availability of the extract, which are key factors in applications that require controlled delivery and enhanced therapeutic efficacy of the extract.

In vitro Drug Release Study

Owing to the presence of numerous bioactive phytoconstituents in plants, it is challenging to precisely quantify all of these components. Therefore, in the present study, a single key bioactive compound was selected as a reference for the analysis of quercetin, which was identified as the primary constituent in the *Sonchus arvensis* L. extract and was employed as the standard compound for evaluating the dissolution study. The concentrations of the active constituents were determined using a quercetin calibration curve. The dissolution profiles of both the crude plant extract and

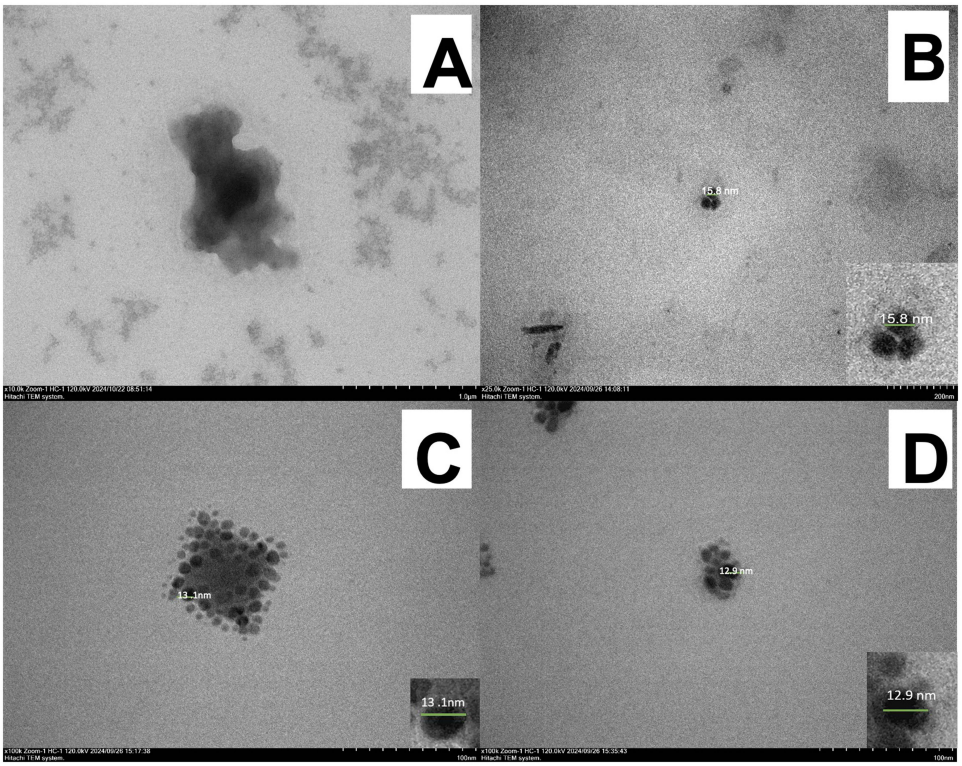

Figure 5 TEM analysis of Sonchus extract and three NS formulations shows the morphology of spherical shape of NSs and Sonchus extract (**A**) Sa-Extract, (**B**) NS-Sa-1:1, (**C**) NS-Sa-2:1, (**D**) NS-Sa-4:1.

nanosuspensions were analyzed in a phosphate buffer solution (pH 7.4) as the dissolving medium. The results indicated that the release of both the plant extract and nanosuspensions increased over time, with a significant difference in the release rates between the two. According to theoretical principles, the in vitro release rate of a drug can be enhanced by decreasing its particle size, thereby increasing its surface area.

The slower release of the crude extract can be attributed to its larger particle size, lack of stabilization, and reduced solubility, which hinder its dissolution.

The enhanced release rate of NS-Sa-4:1 demonstrated the advantages of nanosuspension technology, such as improved particle size reduction, stability, and solubility, achieved in the presence of a high concentration of Tween 80.

The in vitro release study clearly showed that NS-Sa-4:1 outperformed the crude extract in terms of release rate and overall drug release. The superior performance of NS-Sa-4:1 can be attributed to its smaller and more stable particle size, uniform morphology, as observed in the TEM analysis, and better long-term stability. The higher concentration of Tween 80 effectively enhanced drug dissolution and release, making it the best candidate for improving therapeutic efficacy.

The crude extract exhibited a slower and less regulated release profile than the nanosuspension formulations, achieving a cumulative release of approximately 28.9% over 24 h. In contrast, NS-Sa-4:1 displayed a markedly faster and more sustained release profile, reaching a cumulative release of approximately 92.51% within the same period (Figure 6).

Antioxidant Profile Based on Percentage of Inhibition and IC_{50} Values

The analysis of IC_{50} values, derived from the inhibition percentages presented in Figure 7, revealed that NS-Sa-4:1 demonstrated the highest antioxidant activity among the tested formulations, exhibiting efficacy similar to that of ascorbic acid, a standard antioxidant. The superior antioxidant capacity of this formulation is attributed to its reduced particle size, enhanced stability, and uniform particle distribution, which differentiates it from both the raw extract and other nanosuspension formulations. These results highlight the potential of nanosuspension technology for improving the antioxidant properties of bioactive compounds.

Figure 7 illustrates the percentage inhibition of various formulations of *Sonchus arvensis* L. extract (Sa-Extract, NS-Sa-1:1, NS-Sa-2:1, and NS-Sa-4:1) at different concentrations (50, 100, 150, 200, and 250 ppm) compared to ascorbic acid as the control. This comparison underscores the differences in antioxidant efficacy between the raw extract, nanosuspension formulations, and standard antioxidant.

At all concentrations, ascorbic acid consistently demonstrated the highest percentage of inhibition, with statistical significance ($P < 0.01$, denoted by **), reaffirming its superior antioxidant activity. Among the tested formulations, the nanosuspension NS-Sa-4:1 generally exhibited the highest percentage of inhibition, although it remained significantly

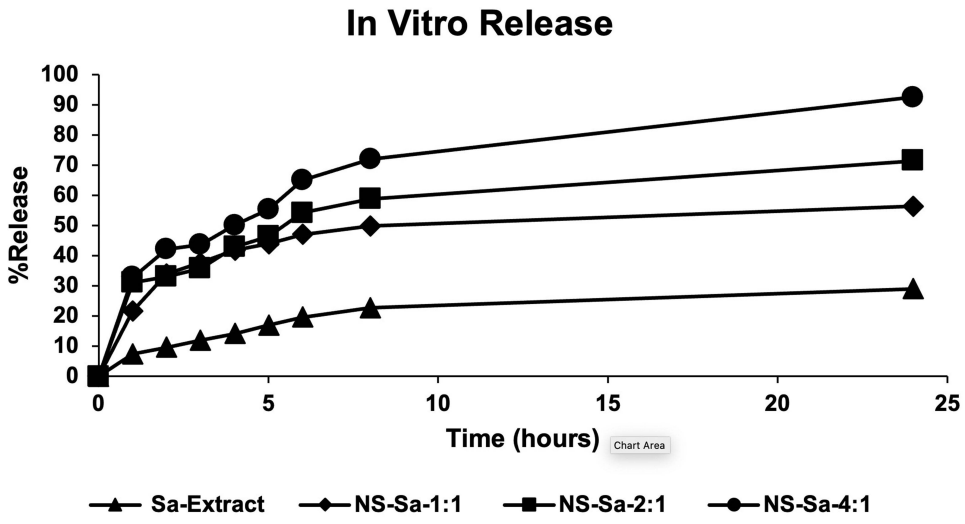

Figure 6 In vitro release of crude extract and nanosuspension over 24 hrs.

https://doi.org/10.2147/IJN.S511602

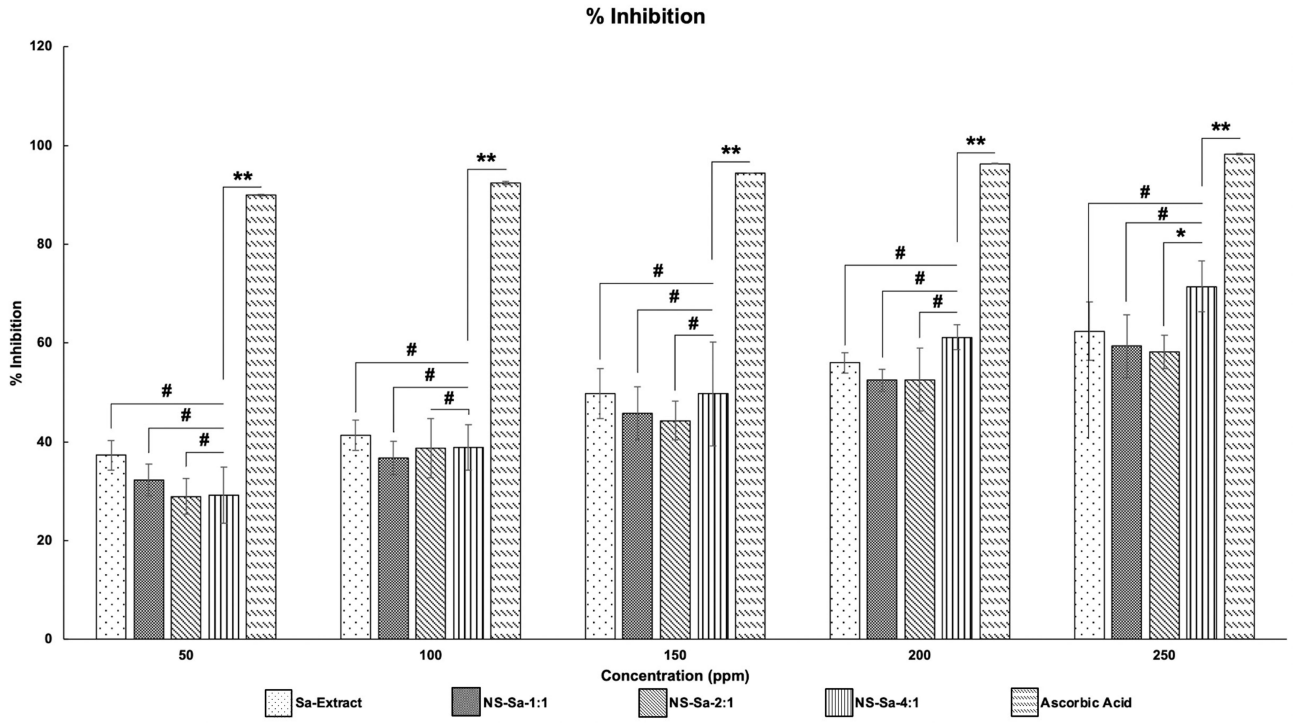

Figure 7 Percentage DPPH inhibition of various formulations of *Sonchus arvensis* L. extract, including the raw extract (Sa-Extract), nanosuspensions prepared with different polymer ratios (NS-Sa-1:1, NS-Sa-2:1, NS-Sa-4:1), and ascorbic acid as a control, at concentrations of 50, 100, 150, 200, and 250 ppm. Data are presented as mean ± standard deviation (SD). ** indicates a significant difference compared to all other groups at $P < 0.01$; *indicates $P < 0.05$; and #denotes non-significant differences ($P > 0.05$).

lower than ascorbic acid at all concentrations ($P < 0.01$). These findings suggest that while nanosuspension technology improves antioxidant activity, it does not reach the efficacy level of the standard.

The raw extract (Sa-Extract) exhibited the lowest percentage of inhibition across all concentrations, emphasizing the limitations of the unprocessed form. Comparisons among the nanosuspension formulations (NS-Sa-1:1, NS-Sa-2:1, and NS-Sa-4:1) revealed no significant differences in their inhibition percentages ($P > 0.05$, denoted by #), indicating comparable antioxidant efficacies among these formulations. This similarity suggests that while nanosuspension technology enhances activity over the raw extract, the polymer ratio within the tested range has a limited impact on antioxidant potential.

At higher concentrations (200 and 250 ppm), all nanosuspension formulations showed increased percentage inhibition, with NS-Sa-4:1 exhibiting a slight superiority. However, the differences among the nanosuspension formulations were not statistically significant ($P > 0.05$). Despite these improvements, the percentage inhibition for all formulations consistently remained significantly lower than that of ascorbic acid ($P < 0.05$ or $P < 0.01$), reinforcing the unmatched potency of the standard.

Further analysis of IC_{50} values, derived from the inhibition percentages presented in Figure 8, reveals that NS-Sa-4:1 demonstrates the highest antioxidant activity among the tested formulations. However, its activity was significantly lower than that of ascorbic acid, which was the positive control used in this experiment. The enhanced antioxidant capacity of this formulation can be attributed to its reduced particle size, improved stability, and uniform particle distribution. These properties likely differentiate it from both the raw extract and other nanosuspension formulations. Overall, the findings underscore the potential of nanosuspension technology to significantly improve the antioxidant properties of bioactive compounds, making it a promising strategy for enhancing the efficacy of natural extracts.

Antihyperlipidemic Based on Percentage of Inhibition and IC_{50} Values

Figure 9 illustrates the percentage of cholesterol inhibition of various formulations, including Sa-Extract (raw extract of *Sonchus arvensis* L.), nanosuspensions (NS-Sa-1:1, NS-Sa-2:1, and NS-Sa-4:1), and simvastatin as a reference standard,

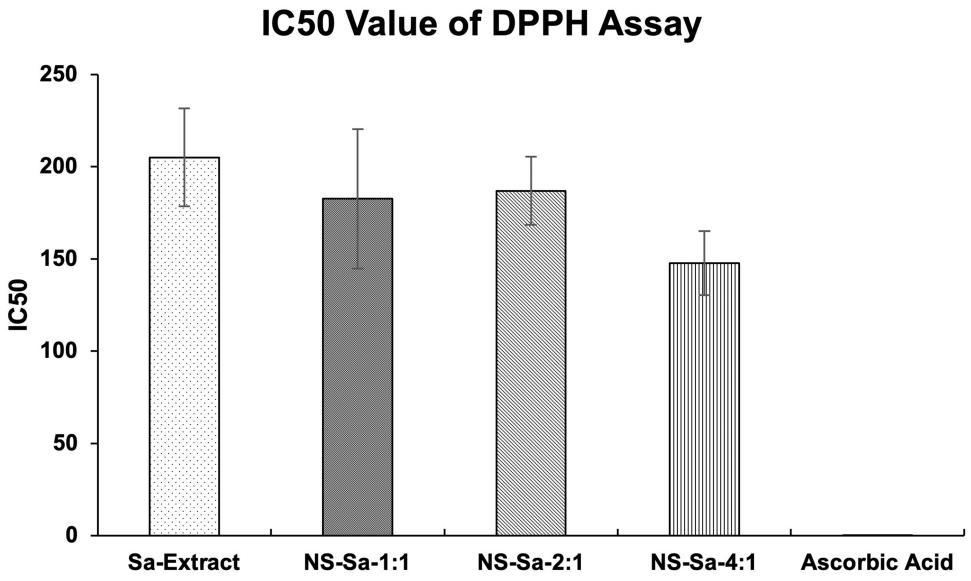

Figure 8 IC_{50} Values of DPPH antioxidant assay.

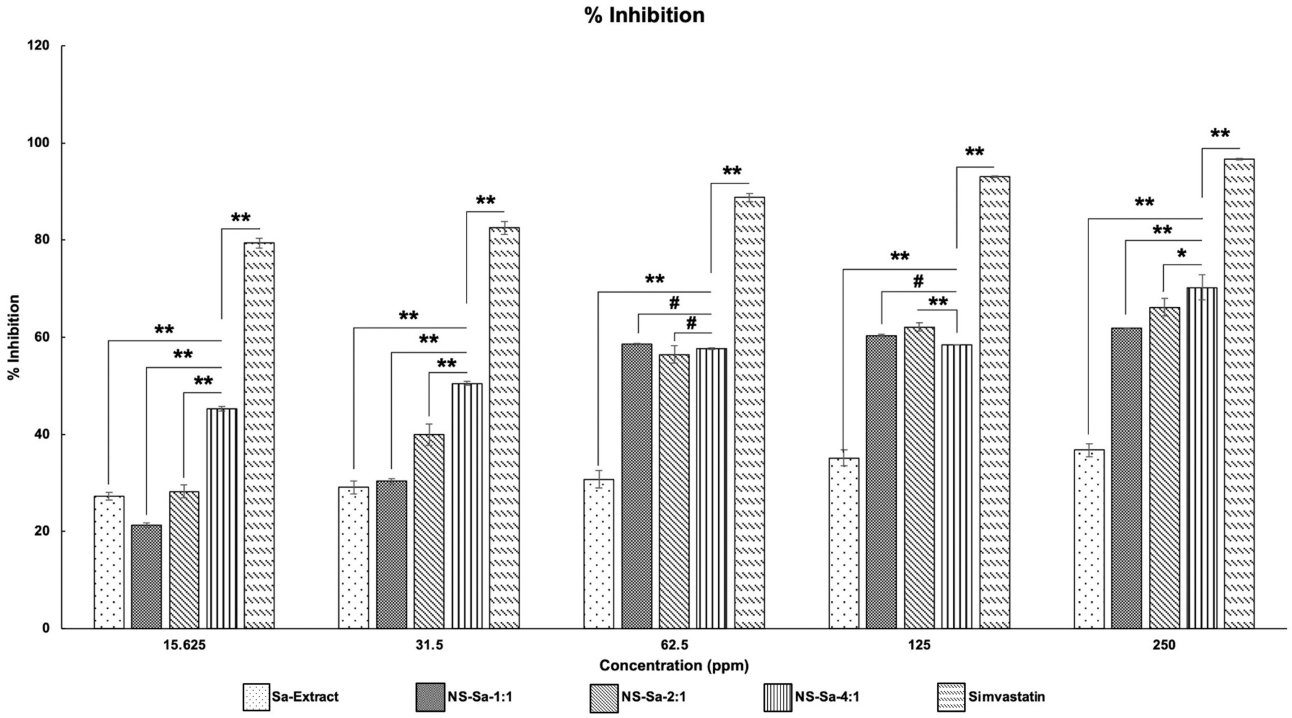

Figure 9 Percentage cholesterol inhibition of Sa-Extract (*Sonchus arvensis* L. raw extract), nanosuspensions (NS-Sa-1:1, NS-Sa-2:1, and NS-Sa-4:1), and Simvastatin as a standard at various concentrations (15.625, 31.5, 62.5, 125, and 250 ppm). Simvastatin consistently exhibited the highest percentage inhibition at all concentrations, with statistically significant differences ($P < 0.01$, denoted by **). Among the nanosuspensions, NS-Sa-4:1 showed the highest inhibition percentage, particularly at higher concentrations, while differences among nanosuspensions were not statistically significant at most concentrations ($P > 0.05$, denoted by #). At 250 ppm, NS-Sa-4:1 demonstrated a slight but significant improvement over NS-Sa-2:1 ($P < 0.05$, denoted by *). Error bars represent standard deviation (SD).

across concentrations ranging from 15.625 to 250 ppm. The results highlighted a consistent pattern of performance among the formulations, with simvastatin demonstrating superior inhibitory activity at all concentrations ($P < 0.01$, denoted by **).

https://doi.org/10.2147/IJN.S511602

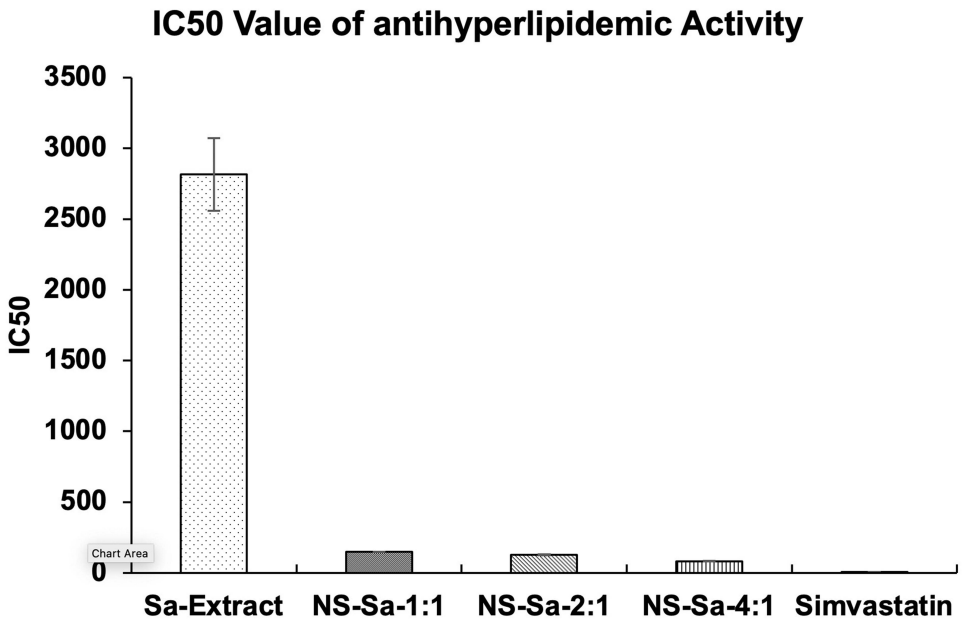

Figure 10 IC_{50} Values of antihyperlipidemic activity.

Among the nanosuspension formulations, NS-Sa-4:1 consistently exhibited the highest percentage of inhibition, followed by NS-Sa-2:1 and NS-Sa-1:1. At lower concentrations (15.625 and 31.5 ppm), the nanosuspensions displayed significantly higher inhibition percentages than the Sa-Extract ($P < 0.01$), yet all remained significantly less effective than simvastatin. At 62.5 ppm, NS-Sa-4:1 showed a slightly higher inhibition percentage than NS-Sa-2:1 and NS-Sa-1:1; however, these differences were not statistically significant ($P > 0.05$, denoted by #).

As the concentration increased (125 and 250 ppm), the percentage inhibition of all nanosuspension formulations improved markedly. NS-Sa-4:1 showed significantly higher inhibition than the Sa extract at both concentrations ($P < 0.01$). However, at 125 ppm, no significant differences were observed among the nanosuspensions ($P > 0.05$). At 250 ppm, NS-Sa-4:1 demonstrated a slight but statistically significant improvement over NS-Sa-2:1 ($P < 0.05$, denoted by *). Despite these improvements, all nanosuspension formulations remained significantly less effective than simvastatin ($P < 0.01$).

The analysis of IC_{50} values (Figure 10), derived from the percentage inhibition data, further illustrates the relative performance of the formulations. IC_{50} values represent the concentration required to achieve 50% inhibition, with lower values indicating greater efficacy. Among the nanosuspensions, NS-Sa-4:1 demonstrated the most pronounced antihyperlipidemic effect, approaching the efficacy of simvastatin. This enhanced performance is attributed to the smaller particle size, improved stability, and uniform particle distribution, which collectively enhance pharmaceutical availability and lipid interaction.

Overall, these findings underscore the therapeutic potential of nanosuspension technology in enhancing the efficacy of lipid-lowering agents, particularly when applied to bioactive compounds such as *Sonchus arvensis L*. The superior performance of NS-Sa-4:1 highlights the importance of optimizing the formulation to improve the clinical potential of natural extracts.

Discussion

This study highlights the use of a green synthetic approach for *Sonchus arvensis* L., marking a substantial step in sustainable nanotechnology. By eliminating the need for hazardous chemicals, this method aligns with global environmental initiatives and emphasizes the importance of eco-friendly practices in drug delivery systems.[37] Notably, this approach leverages the inherent bioactivity of *Sonchus arvensis* L., effectively bridging the gap between traditional

medicinal approaches and advanced nanotechnological innovations.[38] In contrast to conventional methods, which often rely on toxic reagents, this strategy offers an environmentally safe alternative without compromising therapeutic efficacy.

This study breaks new ground by employing a *Sonchus arvensis* L. nanosuspension for antihyperlipidemic activity, offering an innovative, natural, and sustainable solution for managing lipid levels. The nanoprecipitation technique used in this study ensured uniform particle distribution, reduced synthesis time, and enhanced nanosuspension stability. These results were corroborated by Transmission Electron Microscopy (TEM) and zeta potential analysis, which confirmed the consistent morphology and stability of the formulation.[39]

The results of this study showed that the *Sonchus arvensis* L. nanosuspension (NS-Sa-4:1) demonstrated remarkable antihyperlipidemic activity, with therapeutic effects comparable to those of simvastatin ($p < 0.05$). This efficacy can be attributed to its improved pharmaceutical availability and a controlled drug release profile. Furthermore, the nanosuspension exhibited strong antioxidant properties, with an IC_{50} value indicating robust free radical scavenging activity.[40] These findings support the growing evidence of the advantages of nanosuspension formulations in enhancing the solubility and therapeutic performance of bioactive compounds.[41]

The observed improvements in antioxidant and lipid-lowering activities are directly linked to the physicochemical properties of the nanosuspension. The enhanced pharmaceutical availability, controlled drug release profile, and uniform particle distribution achieved through nanoprecipitation contribute to superior therapeutic efficacy. The smaller particle size of nanosuspensions increases the surface area, facilitating better solubility and cellular uptake, which in turn enhances their pharmacological effects. Additionally, the stable zeta potential observed in this study ensured the stability of the formulation, preventing aggregation and preserving its bioactivity. These physicochemical attributes collectively reinforce the potential of nanosuspensions as advanced and sustainable drug delivery systems.

Compared with previous studies, this study highlights the unique benefits of the nanosuspension formulation of *Sonchus arvensis* L. While earlier research predominantly focused on crude plant extracts, encapsulating *Sonchus arvensis* L. phytoconstituents within nanoparticles has demonstrated significantly improved stability and therapeutic potential. Supporting evidence from studies on gemfibrozil and curcumin nanosuspensions further validates the efficacy of nanoprecipitation in enhancing drug performance.[42,43] This approach not only amplifies the lipid-lowering and antioxidant potential of *Sonchus arvensis* L. but also redefines its application in the treatment of hyperlipidemia.

Conclusion

The nanosuspension formulation of *Sonchus arvensis* L. folium has been demonstrated to significantly enhanced its antihyperlipidemic and antioxidant effects compared to the crude extract. The optimized NS-Sa-4:1 formulation exhibited desirable physicochemical properties, including reduced particle size, improved stability, and enhanced solubility, which contributed to its superior in vitro release and bioactivity. These enhancements resulted in antihyperlipidemic and antioxidant effects comparable to those of conventional treatments, such as simvastatin and ascorbic acid, suggesting that nanosuspension technology effectively improves the pharmacokinetic and pharmacodynamic profiles of *S. arvensis* L. The observed improvements in lipid regulation and oxidative stress reduction highlight the potential of this formulation as a viable natural alternative for managing hyperlipidemia and oxidative stress-related disorders. These findings reinforce the therapeutic relevance of nanosuspension-based drug delivery for optimizing the efficacy of herbal bioactives, paving the way for further in vivo studies and potential clinical applications of this approach.

Acknowledgments

The authors extend their appreciation to Northern Border University, Saudi Arabia, for supporting this work through project number (NBU-CRP-2025-2985).

Funding

We would like to thank the Rector of Universitas Padjadjaran for the APC. We also extend our gratitude to the Indonesian Ministry of Higher Education, Science, and Technology for the 2025 doctoral research grant, and to the Libyan Ministry of Higher Education and Elmergib University for the doctoral scholarship.

Disclosure

The authors declare no conflicts of interest in this work.

References

1. Merćep I, Strikić D, Slišković AM, Reiner Ž. New therapeutic approaches in treatment of dyslipidaemia—a narrative review. *Pharmaceuticals*. 2022;15(7):839. doi:10.3390/ph15070839
2. El-Tantawy WH, Temraz A. Natural products for controlling hyperlipidemia. *Arch Physiol Biochem*. 2019;125(2):128–135. doi:10.1080/13813455.2018.1441315
3. Ezeh KJ, Ezeudemba O. Hyperlipidemia: a review of the novel methods for the management of lipids. *Cureus*. 2021;13(7).
4. Fiorentino R, Chiarelli F. Treatment of dyslipidaemia in children. *Biomedicines*. 2021;9(9):1078. doi:10.3390/biomedicines9091078
5. Pacifico L, Bonci E, Di Martino M, et al. A double-blind, placebo-controlled randomized trial to evaluate the efficacy of docosahexaenoic acid supplementation on hepatic fat and associated cardiovascular risk factors in overweight children with nonalcoholic fatty liver disease. *Nutr Metab Cardiovasc Dis*. 2015;25(8):734–741. doi:10.1016/j.numecd.2015.04.003
6. Najmi A, Javed SA, Al Bratty M, Alhazmi HA. Modern approaches in the discovery and development of plant-based natural products and their analogues as potential therapeutic agents. *Molecules*. 2022;27(2):349. doi:10.3390/molecules27020349
7. Dzobo K. The role of natural products as sources of therapeutic agents for innovative drug discovery. *Compr Pharmacol*. 2022:408.
8. Li XM, Yang PL. Research progress of Sonchus species. *Int J Food Prop*. 2018;21(1):147–157. doi:10.1080/10942912.2017.1415931
9. Winarto IWP, Karyasari T. *Tempuyung; Tanaman Penghancur Batu Ginjal*. AgroMedia; 2004.
10. Tandi J, Sutrisna INE, Pratiwi M, Handayani TW. Potential test of nephropathy Sonchus arvensis L. leaves on male rats (Rattus norvegicus) diabetes mellitus. *Pharmacogn J*. 2020;12(5):1115–1120. doi:10.5530/pj.2020.12.158
11. Bhalani DV, Nutan B, Kumar A, Singh Chandel AK. Bioavailability enhancement techniques for poorly aqueous soluble drugs and therapeutics. *Biomedicines*. 2022;10(9):2055. doi:10.3390/biomedicines10092055
12. Katona MT, Nagy-Katona L, Szabó R, Borbás E, Tonka-Nagy P, Takács-Novák K. Multi-compartmental dissolution method, an efficient tool for the development of enhanced bioavailability formulations containing poorly soluble acidic drugs. *Pharmaceutics*. 2023;15(3):753. doi:10.3390/pharmaceutics15030753
13. Du Y, Yuan X. Coupled hybrid nanoparticles for improved dispersion stability of nanosuspensions: a review. *J Nanopart Res*. 2020;22(9):261. doi:10.1007/s11051-020-04991-8
14. Perrin L, Pajor-Swierzy A, Magdassi S, Kamyshny A, Ortega F, Rubio RG. Evaporation of nanosuspensions on substrates with different hydrophobicity. *ACS Appl Mater Interfaces*. 2018;10(3):3082–3093. doi:10.1021/acsami.7b15743
15. Liu T, Yu X, Yin H, Möschwitzer JP. Advanced modification of drug nanocrystals by using novel fabrication and downstream approaches for tailor-made drug delivery. *Drug Deliv*. 2019;26(1):1092–1103. doi:10.1080/10717544.2019.1682721
16. Lynnerup JT, Eriksen JB, Bauer-Brandl A, Holsæter AM, Brandl M. Insight into the mechanism behind oral bioavailability-enhancement by nanosuspensions through combined dissolution/permeation studies. *Eur J Pharm Sci*. 2023;184:106417. doi:10.1016/j.ejps.2023.106417
17. Azimullah S, Sudhakar CK, Kumar P, et al. Nanosuspensions as a promising approach to enhance bioavailability of poorly soluble drugs: an update. *J Drug Delivery Ther*. 2019;9(2):574–582. doi:10.22270/jddt.v9i2.2436
18. Khandbahale SV. A review-Nanosuspension technology in drug delivery system. *Asian J Pharm Res*. 2019;9(2):130–138. doi:10.5958/2231-5691.2019.00021.2
19. Dewangan HK. The emerging role of nanosuspensions for drug delivery and stability. *Curr Nanomed*. 2021;11(4):213–223. doi:10.2174/2468187312666211222123307
20. Dewi NP, Alaydrus S, Lestari WA, et al. Suspension of Sonchus arvensis L. leaves ethanolic extract affects blood creatinine and urea levels in streptozotocin-induced wistar male rats. *Sci Pharm*. 2024;3(1):18–23.
21. Kemenkes RI. Farmakope Herbal Indonesia Edisi II. *Jakarta Kementrian Kesehat RI*. 2017.
22. Zhang T, Li X, Xu J, Shao J, Ding M, Shi S. Preparation, characterization, and evaluation of breviscapine nanosuspension and its freeze-dried powder. *Pharmaceutics*. 2022;14(5):923. doi:10.3390/pharmaceutics14050923
23. Chavhan R. Nanosuspensions: enhancing drug bioavailability through nanonization. In: *Annales Pharmaceutiques Françaises*. Elsevier; 2024.
24. Hussain A, Attique F, Naqvi SAR, et al. Nanoformulation of curcuma longa root extract and evaluation of its dissolution potential. *ACS Omega*. 2022. doi:10.1021/acsomega.2c06258
25. Thenge RR, Patel A, Mehetre G. Nanocrystals in the drug delivery system. In: *Photophysics and Nanophysics in Therapeutics*. Elsevier; 2022:443–454.
26. Elmowafy M, Shalaby K, Al-Sanea MM, et al. Influence of stabilizer on the development of luteolin nanosuspension for cutaneous delivery: an in vitro and in vivo evaluation. *Pharmaceutics*. 2021;13(11):1812. doi:10.3390/pharmaceutics13111812
27. Shaikh F, Patel M, Patel V, et al. Formulation and optimization of cilnidipine loaded nanosuspension for the enhancement of solubility, dissolution and bioavailability. *J Drug Deliv Sci Technol*. 2022;69:103066. doi:10.1016/j.jddst.2021.103066
28. Casula L, Sinico C, Valenti D, et al. Delivery of beclomethasone dipropionate nanosuspensions with an electronic cigarette. *Int J Pharm*. 2021;596:120293. doi:10.1016/j.ijpharm.2021.120293
29. Suo Z, Sun Q, Peng X, et al. Lentinan as a natural stabilizer with bioactivities for preparation of drug–drug nanosuspensions. *Int J Biol Macromol*. 2021;184:101–108. doi:10.1016/j.ijbiomac.2021.06.056
30. Rasul A, Imran Khan M, Ur Rehman M, et al. In vitro characterization and release studies of combined nonionic surfactant-based vesicles for the prolonged delivery of an immunosuppressant model drug. *Int J Nanomed*. 2020;Volume 15:7937–7949. doi:10.2147/IJN.S268846
31. Raisawati T, Melati M, Aziz SA, Rafi M. Total flavonoid content and antioxidant activity in leaves of ten cultivated Sonchus arvensis L. accessions. *Plant Arch*. 2020;20(1).
32. Luhurningtyas FP, Vifta RL, Syarohmawati N, Candra MA. Cholestrol lowering effect of chitosan nanoparticles using Parijoto fruits extract. *J Farm Sains Dna Komunitas*. 2020;17(2):102–111.

33. Muharni M, Julinar J, Yohandini H, Fitrya F, Melati R. In vitro anti-cholesterol and anti-hypertensive activity of stem bark the Flacourtia rukam. In: *IOP Conference Series: Earth and Environmental Science*. Vol. 713. IOP Publishing; 2021:12056.
34. Zhao Y, Wang Z, Zhang W, Jiang X. Adsorbed Tween 80 is unique in its ability to improve the stability of gold nanoparticles in solutions of biomolecules. *Nanoscale*. 2010;2(10):2114–2119. doi:10.1039/c0nr00309c
35. Clogston JD, Patri AK. Zeta potential measurement. In: *Characterization of Nanoparticles Intended for Drug Delivery*. Springer; 2011:63–70.
36. Patra JK, Das G, Fraceto LF, et al. Nano based drug delivery systems: recent developments and future prospects. *J Nanobiotechnology*. 2018;16 (1):1–33. doi:10.1186/s12951-017-0328-8
37. Ahmad S, Munir S, Zeb N, et al. Green nanotechnology: a review on green synthesis of silver nanoparticles—An ecofriendly approach. *Int J Nanomed*. 2019;Volume 14:5087–5107. doi:10.2147/IJN.S200254
38. Suwartiny NL, Rafi M, Rohaeti E. Traditional use, phytochemical composition, and biological activities of Sonchus arvensis L. *Indones J Pharmacy*. 2022;33(4).
39. Lammari N, Louaer O, Meniai AH, Elaissari A. Encapsulation of essential oils via nanoprecipitation process: overview, progress, challenges and prospects. *Pharmaceutics*. 2020;12(5):431. doi:10.3390/pharmaceutics12050431
40. Wei L, Wu H, Wang X, Wen L, Cui B, Cheng Y. Comprehensive review of plant-derived anti-hyperlipidemia peptides: production, anti-hyperlipidemia mechanism, and structure-activity relationship study. *Food Chem*. 2024;461:140715. doi:10.1016/j.foodchem.2024.140715
41. Ahmed HM, Nabavi S, Behzad S. Herbal drugs and natural products in the light of nanotechnology and nanomedicine for developing drug formulations. *Mini Rev Med Chem*. 2021;21(3):302–313. doi:10.2174/1389557520666200916143240
42. Patel K, Patil J, Girase T, Tatiya A, Patil D. An overview of the applications of gemfibrozil Nano-formulation in hyperlipidemia. *Mater Proc*. 2023;14(1):45.
43. Elbaz NM, Tatham LM, Owen A, Rannard S, McDonald TO. Redispersible nanosuspensions as a plausible oral delivery system for curcumin. *Food Hydrocoll*. 2021;121:107005. doi:10.1016/j.foodhyd.2021.107005